

SEQUENCE LISTING

<110> Koide, Shohei

<120> METHOD OF IDENTIFYING POLYPEPTIDE MONOBODIES WHICH BIND TO TARGET PROTEINS AND USE THEREOF

<130> 176/60901

<140> 10/006,760

<141> 2001-11-19

<150> 60/249,756

<151> 2000-11-17

<160> 73

<170> PatentIn Ver. 2.1

<210> 1

<211> 308

<212> DNA

<213> Homo sapiens

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<210> 2

<211> 96

<212> PRT

<213> Homo sapiens

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Pro Thr Ser Leu Leu Ile Ser Trp Asp Ala Pro Ala Val Thr Val Arg

Tyr Tyr Arg Ile Thr Tyr Gly Glu Thr Gly Gly Asn Ser Pro Val Gln
35 40 45

Glu Phe Thr Val Pro Gly Ser Lys Ser Thr Ala Thr Ile Ser Gly Leu 50 55 60

Lys Pro Gly Val Asp Tyr Thr Ile Thr Val Tyr Ala Val Thr Gly Arg
65 70 75 80

Gly Asp Ser Pro Ala Ser Ser Lys Pro Ile Ser Ile Asn Tyr Arg Thr 85 90 95

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<212> PRT

<213> Artificial Sequence

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Pro Thr Ser Leu Leu Ile Ser Trp Asp Ala Pro Ala Val Thr Val Arg
20 25 30

Tyr Tyr Arg Ile Thr Tyr Gly Glu Thr Gly Gly Asn Ser Pro Val Gln
35 40 45

Glu Phe Thr Val Pro Gly Ser Lys Ser Thr Ala Thr Ile Ser Gly Leu
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Lys Pro Gly Val Asp Tyr Thr Ile Thr Val Tyr Ala Val Thr Gly Arg
65 70 75 . 80

Gly Asp Ser Pro Ala Ser Ser Lys Pro Ile Ser Ile Asn Tyr Arg Thr 85 90 95

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accatcactg tatacgctgt tactggcnnk nnknnknnkn nknnknnktc caagccaatc 300
tcgattaact accgtaccag tggtaccggt ggttcccctc caaaaaagaa gagaaaggta 360
gctggtatca ataaagatat cgaggagtgc aatgccatca ttgagcagtt tatcgactac 420
ctgcgcaccg gacaggagat gccgatggaa atggcggatc aggcgattaa cgtggtgccg 480
ggcatgacgc cgaaaaccat tcttcacgcc gggccgccga tccagcctga ctggctgaaa 540
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 <223> Xaa at any position can be any amino acid
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 <222> (90)..(96)
 <223> Xaa at any position can be any amino acid
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                                  25
              20
 Ser Trp Asp Ala Pro Xaa Xaa Xaa Xaa Tyr Tyr Arg Ile Thr Tyr
 Gly Glu Thr Gly Gly Asn Ser Pro Val Glu Phe Thr Val Pro Gly
                     . 55
 Ser Lys Ser Thr Ala Thr Ile Ser Gly Leu Lys Pro Gly Val Asp Tyr
                      70
                                          75
  65
 Thr Ile Thr Val Tyr Ala Val Thr Gly Xaa Xaa Xaa Xaa Xaa Xaa Xaa
                  85
                                      90
 Ser Lys Pro Ile Ser Ile Asn Tyr Arg Thr Ser Gly Thr Gly Gly Ser.
             100
                                 105 .
                                                     110
 Pro Pro Lys Lys Arg Lys Val Ala Gly Ile Asn Lys Asp Ile Glu
         115
 Glu Cys Asn Ala Ile Ile Glu Gln Phe Ile Asp Tyr Leu Arg Thr Gly
                         135
                                             140
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Gln Glu Met Pro Met Glu Met Ala Asp Gln Ala Ile Asn Val Val Pro 155 160 145 150 Gly Met Thr Pro Lys Thr Ile Leu His Ala Gly Pro Pro Ile Gln Pro 165 170 Asp Trp Leu Lys Ser Asn Gly Phe His Glu Ile Glu Ala Asp Val Asn 180 185 190 Asp Thr Ser Leu Leu Leu Ser Gly Asp 200 195 <210> 6 <211> 96 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FNfn10 polypeptide monobody <220> <221> UNSURE <222> (28)..(32) <223> Xaa at any position can be any amino acid <220> <221> UNSURE <222> (80)..(86) <223> Xaa at any position can be any amino acid Met Gln Val Ser Asp Val Pro Thr Asp Leu Glu Val Val Ala Ala Thr . 10 15 5 Pro Thr Ser Leu Leu Ile Ser Trp Asp Ala Pro Xaa Xaa Xaa Xaa 20 Tyr Tyr Arg Ile Thr Tyr Gly Glu Thr Gly Gly Asn Ser Pro Val Gln 40 45 35 Glu Phe Thr Val Pro Gly Ser Lys Ser Thr Ala Thr Ile Ser Gly Leu 5**5** 50

Lys Pro Gly Val Asp Tyr Thr Ile Thr Val Tyr Ala Val Thr Gly Xaa

75

70

Xaa Xaa Xaa Xaa Xaa Ser Lys Pro Ile Ser Ile Asn Tyr Arg Thr 85 90 95

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gatcaggcga ttaacgtggt gccgggcatg acgccgaaaa ccattcttca cgccgggccg 240
ccgatccagc ctgactggct gaaatcgaat ggttttcatg aaattgaagc ggatgttaac 300
gataccagcc tettgetgag tggagatgcc tecaagettg gtaccgaget eggatetatg 360
caggtttctg atgttccgac cgacctggaa gttgttgctg cgaccccgnn snnsnnsnns 420
nnsnnsnnsa ctagcctgct gatcagctgg gatgctcctg cagttaccgt gcgttattac 480
cgtatcacgt acggtgaaac cggtggtaac tccccggttc aggaattcac tgtacctggt 540
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 fusion protein

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<222> (137)..(143)

<223> Xaa at any position can be any amino acid

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1 5 10 15

Ala Met Gly Ala Pro Pro Lys Lys Lys Arg Lys Val Ala Gly Ile Asn
20 25 30

Lys Asp Ile Glu Glu Cys Asn Ala Ile Ile Glu Gln Phe Ile Asp Tyr 35 40 45

Leu Arg Thr Gly Gln Glu Met Pro Met Glu Met Ala Asp Gln Ala Ile 50 55 60

Asn Val Val Pro Gly Met Thr Pro Lys Thr Ile Leu His Ala Gly Pro 65 70 75 80

Pro Ile Gln Pro Asp Trp Leu Lys Ser Asn Gly Phe His Glu Ile Glu
85 90 95

Ala Asp Val Asn Asp Thr Ser Leu Leu Leu Ser Gly Asp Ala Ser Lys
100 ... 105. 110

Leu Gly Thr Glu Leu Gly Ser Met Gln Val Ser Asp Val Pro Thr Asp 115 120 125

Leu Glu Val Val Ala Ala Thr Pro Xaa Xaa Xaa Xaa Xaa Xaa Xaa Thr 130 135 140

Ser Leu Leu Ile Ser Trp Asp Ala Pro Ala Val Thr Val Arg Tyr Tyr

145 150 155 160

Arg Ile Thr Tyr Gly Glu Thr Gly Gly Asn Ser Pro Val Gln Glu Phe 165 170 175

Thr Val Pro Gly Ser Lys Ser Thr Ala Thr Ile Ser Gly Leu Lys Pro 180 185 190

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 polypeptide monobody

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<223> Xaa at any position can be any amino acid

<400> 9

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Pro Xaa Xaa Xaa Xaa Xaa Xaa Thr Ser Leu Leu Ile Ser Trp Asp 20 25 30

Ala Pro Ala Val Thr Val Arg Tyr Tyr Arg Ile Thr Tyr Gly Glu Thr 35 40 45

Gly Gly Asn Ser Pro Val Gln Glu Phe Thr Val Pro Gly Ser Lys Ser 50 55 60

Thr Ala Thr Ile Ser Gly Leu Lys Pro Gly Val Asp Tyr Thr Ile Thr
65 70 75 80

Val Tyr Ala Val Thr Gly Arg Gly Asp Ser Pro Ala Ser Ser Lys Pro 85 90 95

Ile Ser Ile Asn Tyr Arg Thr 100

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agcggcctga aaccgggtgt cgactatacc atcactgtat acgctgttac tggcnnknnk 600
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 fusion protein

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<222> (199)..(205)

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Met Gly Lys Pro Ile Pro Asn Pro Leu Leu Gly Leu Asp Ser Thr Gln
1 5 10 15

Ala Met Gly Ala Pro Pro Lys Lys Lys Arg Lys Val Ala Gly Ile Asn 20 25 30

Lys Asp Ile Glu Glu Cys Asn Ala Ile Ile Glu Gln Phe Ile Asp Tyr 35 . 40 . 45

Leu Arg Thr Gly Gln Glu Met Pro Met Glu Met Ala Asp Gln Ala Ile 50 . 55 60

Asn Val Val Pro Gly Met Thr Pro Lys Thr Ile Leu His Ala Gly Pro 65 70 75 80

Pro Ile Gln Pro Asp Trp Leu Lys Ser Asn Gly Phe His Glu Ile Glu 85 90 95

Ala Asp Val Asn Asp Thr Ser Leu Leu Leu Ser Gly Asp Ala Ser Lys
100 105 110

Leu Gly Thr Glu Leu Gly Ser Met Gln Val Ser Asp Val Pro Thr Asp

115 120 125

Leu Glu Val Val Ala Ala Thr Pro Thr Ser Leu Leu Ile Ser Trp Asp 130 135 140

Gly Gly Asn Ser Pro Val Gln Glu Phe Thr Val Pro Gly Ser Lys Ser 165 170 175

Thr Ala Thr Ile Ser Gly Leu Lys Pro Gly Val Asp Tyr Thr Ile Thr
180 185 . 190

Val Tyr Ala Val Thr Gly Xaa Xaa Xaa Xaa Xaa Xaa Xaa Ser Lys Pro 195 200 205

Ile Ser Ile Asn Tyr Arg Thr 210 215

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<213> Artificial Sequence . . .

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 polypeptide monobody

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<222> (80)..(85)

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Pro Thr Ser Leu Leu Ile Ser Trp Asp Ala Pro Xaa Xaa Xaa Xaa Xaa 20 25 30

Tyr Tyr Arg Ile Thr Tyr Gly Glu Thr Gly Gly Asn Ser Pro Val Gln
35 40 45

Glu Phe Thr Val Pro Gly Ser Lys Ser Thr Ala Thr Ile Ser Gly Leu 50 55 60

Lys Pro Gly Val Asp Tyr Thr Ile Thr Val Tyr Ala Val Thr Gly Xaa 65 70 75 80

Xaa Xaa Xaa Xaa Xaa Ser Lys Pro Ile Ser Ile Asn Tyr Arg Thr 85 90 95

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<223> N at positions 601 and 602 can be A, C, T, or G

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      or G
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19

<222> (618) <223> K at position 618 can be G or T <220> <221> unsure <222> (621) <223> K at position 621 can be G or T <220> <221> unsure <222> (624) <223> K at position 624 can be G or T <220> <221> unsure <222> (627) <223> K at position 627 can be G or T <220> <221> unsure <222> (630) <223> K at position 630 can be G or T <220> <221> unsure <222> (633) <223> K at position 633 can be G or T <220> <221> unsure <222> (636) <223> K at position 636 can be G or T <220> <221> unsure <222> (639) <223> K at position 639 can be G or T <220> <221> unsure <222> (642) <223> K at position 642 can be G or T <400> 13 atgggtaagc ctatccctaa ccctctcctc ggtctcgatt ctacacaagc tatgggtgct 60 cctccaaaaa agaagagaaa ggtagctggt atcaataaag atatcgagga gtgcaatgcc 120 atcattgagc agtttatcga ctacctgcgc accggacagg agatgccgat ggaaatggcg 180 gatcaggcga ttaacgtggt gccgggcatg acgccgaaaa ccattcttca cgccgggccg 240

<210> 14

<211> 223

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: B42-FNfn10 fusion protein

<220>

<221> UNSURE

<222> (199)..(214)

<223> Xaa at positions 199, 200, 201, 202, 203, 204, 205, 206, 207, 208, 209, 210, 211, 212, 213, and 214 can be any amino acid

<400> 14

Met Gly Lys Pro Ile Pro Asn Pro Leu Leu Gly Leu Asp Ser Thr Gln
1 5 10 15

Ala Met Gly Ala Pro Pro Lys Lys Lys Arg Lys Val Ala Gly Ile Asn 20 25 30

Lys Asp Ile Glu Glu Cys Asn Ala Ile Ile Glu Gln Phe Ile Asp Tyr 35 40 45

Leu Arg Thr Gly Gln Glu Met Pro Met Glu Met Ala Asp Gln Ala Ile 50 55. 60

Asn Val Val Pro Gly Met Thr Pro Lys Thr Ile Leu His Ala Gly Pro 65 70 75 80

Pro Ile Gln Pro Asp Trp Leu Lys Ser Asn Gly Phe His Glu Ile Glu 85 90 95

Ala Asp Val Asn Asp Thr Ser Leu Leu Leu Ser Gly Asp Ala Ser Lys
100 105 110

Leu Gly Thr Glu Leu Gly Ser Met Arg Val Ser Asp Val Pro Arg Asp 115 120 125

Leu Glu Val Val Ala Ala Thr Pro Thr Ser Leu Leu Ile Ser Trp Asp . 130 135 140

Ala Pro Ala Val Thr Val Arg Tyr Tyr Arg Ile Thr Tyr Gly Glu Thr 145 150 155 160

Gly Gly Asn Ser Pro Val Gln Glu Phe Thr Val Pro Gly Ser Lys Ser 165 170 175

Thr Ala Thr Ile Ser Gly Leu Lys Pro Gly Val Asp Tyr Thr Ile Thr 180 185 190

Xaa Xaa Xaa Xaa Xaa Xaa Lys Pro Ile Ser Ile Asn Tyr Arg Thr 210 215 220

<210> 15

<211> 104

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: FNfn10 polypeptide monobody

<220>

<221> UNSURE

<222> (80)..(95)

<223> Xaa at positions 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, and 95 can be any amino acid

<400> 15

Met Arg Val Ser Asp Val Pro Arg Asp Leu Glu Val Val Ala Ala Thr
1 5 10 15

Pro Thr Ser Leu Leu Ile Ser Trp Asp Ala Pro Ala Val Thr Val Arg 20 25 30

Tyr Tyr Arg Ile Thr Tyr Gly Glu Thr Gly Gly Asn Ser Pro Val Gln
35 40 45

Glu Phe Thr Val Pro Gly Ser Lys Ser Thr Ala Thr Ile Ser Gly Leu 50 55 Lys Pro Gly Val Asp Tyr Thr Ile Thr Val Tyr Ala Val Thr Gly Xaa 70 75 8.5 90 Pro Ile Ser Ile Asn Tyr Arg Thr 100 <210> 16 <211> 663 <212> DNA <213> Artificial Sequence <220> <223> Description of Artificial Sequence: B42-FNfn10 fusion protein coding region <400> 16 atgggtaage ctatecetaa eceteteete ggtetegatt etacacaage tatgggtget 60 cctccaaaaa agaagagaaa ggtagctggt atcaataaag atatcgagga gtgcaatgcc 120 atcattgagc agtttatcga ctacctgcgc accggacagg agatgccgat ggaaatggcg 180 gatcaggcga ttaacgtggt gccgggcatg acgccgaaaa ccattcttca cgccgggccg 240 ccgatccagc ctgactggct gaaatcgaat ggttttcatg aaattgaagc ggatgttaac 300 gataccagec tettgetgag tggagatgee tecaagettg gtaccgaget eggatetatg 360 caggittetg atgiteegae egaeetggaa gitgitgetg egaeeeegae tageetgetg 420 atcagctggg atgctcctgc agttaccgtg cgttattacc gtatcacgta cggtgaaacc 480 ggtggtaact ccccggttca ggaattcact gtacctggtt ccaagtctac tgctaccatc 540 agcggcctga aaccgggtgt cgactatacc atcactgtat acgctgttac tggccgtggt 600 gacageceag egageteeaa gecaateteg attaactace gtaeetagta actegaggea 660 tac 663 <210> 17 <211> 215 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: B42-FNfn10 fusion protein

Met Gly Lys Pro Ile Pro Asn Pro Leu Gly Leu Asp Ser Thr Gln

<400> 17

1 5 10 15

Ala Met Gly Ala Pro Pro Lys Lys Lys Arg Lys Val Ala Gly Ile Asn 20 25 30

Lys Asp Ile Glu Glu Cys Asn Ala Ile Ile Glu Gln Phe Ile Asp Tyr 35 40 45

Leu Arg Thr Gly Gln Glu Met Pro Met Glu Met Ala Asp Gln Ala Ile 50 55 60

Asn Val Val Pro Gly Met Thr Pro Lys Thr Ile Leu His Ala Gly Pro 65 70 75 80

Pro Ile Gln Pro Asp Trp Leu Lys Ser Asn Gly Phe His Glu Ile Glu 85 90 95

Ala Asp Val Asn Asp Thr Ser Leu Leu Leu Ser Gly Asp Ala Ser Lys
100 105 110

Leu Gly Thr Glu Leu Gly Ser Met Gln Val Ser Asp Val Pro Thr Asp 115 120 125

Leu Glu Val Val Ala Ala Thr Pro Thr Ser Leu Leu Ile Ser Trp Asp 130 . 135 140

Ala Pro Ala Val Thr Val Arg Tyr Tyr Arg Ile Thr Tyr Gly Glu Thr 145 150 155 160

Gly Gly Asn Ser Pro Val Gln Glu Phe Thr Val Pro Gly Ser Lys Ser 165 170 175

Thr Ala Thr Ile Ser Gly Leu Lys Pro Gly Val Asp Tyr Thr Ile Thr 180 185 190

Val Tyr Ala Val Thr Gly Arg Gly Asp Ser Pro Ala Ser Ser Lys Pro 195 200 205

Ile Ser Ile Asn Tyr Arg Thr 210 215

<210> 18

<211> 1542

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: lexA-ER(alpha)EF fusion protein

```
<400> 18
atgaaagcgt taacggccag gcaacaagag gtgtttgatc tcatccgtga tcacatcagc 60
cagacaggta tgccgccgac gcgtgcggaa atcgcgcagc gtttggggtt ccgttcccca 120
aacgcggctg aagaacatct gaaggcgctg gcacgcaaag gcgttattga aattgtttcc 180
ggcgcatcac gcgggattcg tctgttgcag gaagaggaag aagggttgcc gctggtaggt 240
cgtgtggctg ccggtgaacc acttctggcg caacagcata ttgaaggtca ttatcaggtc 300
gatccttcct tattcaagcc gaatgctgat ttcctgctgc gcgtcagcgg gatgtcgatg. 360
aaagatatcg gcattatgga tggtgacttg ctggcagtgc ataaaactca ggatgtacgt 420
aacggtcagg tegttgtege aegtattgat gacgaagtta eegttaageg eetgaaaaa 480
cagggcaata aagtcgaact gttgccagaa aatagcgagt ttaaaccaat tgtcgtagat 540
cttcgtcagc agagcttcac cattgaaggg ctggcggttg gggttattcg caacggcgac 600
tggctggaat tcaagcttga gctcggcggc agcggtatga tcaaacgctc taagaagaac 660
agcctggcct tgtccctgac ggccgaccag atggtcagtg ccttgttgga tgctgagccc 720
cccatactct attccgagta tgatcctacc agacccttca gtgaagcttc gatgatgggc 780
ttactgacca acctggcaga cagggagctg gttcacatga tcaactgggc gaagagggtg 840
ccaggetttg tggatttgac cctccatgat caggtccacc ttctagaatg tgcctggcta 900
gagateetga tgattggtet egtetggege teeatggage acceagtgaa getactgttt 960
gctcctaact tgctcttgga caggaaccag ggaaaatgtg tagagggcat ggtggagatc 1020
ttcgacatgc tgctggctac atcatctcgg ttccgcatga tgaatctgca gggagaggag 1080
tttgtgtgcc tcaaatctat tattttgctt aattctggag tgtacacatt tctgtccagc 1140
accetgaagt etetggaaga gaaggaeeat ateeacegag teetggaeaa gateacagae 1200
actttgatcc acctgatggc caaggcaggc ctgaccctgc agcagcagca ccagcggctg 1260
gcccagctcc tcctcatcct ctcccacatc aggcacatga gtaacaaagg catggagcat 1320
ctgtacagca tgaagtgcaa gaacgtggtg cccctctatg acctgctgct ggagatgctg 1380
gacgcccacc gcctacatgc gcccactagc cgtggagggg catccgtgga ggagacggac 1440
caaagccact tggccactgc gggctctact tcatcgcatt ccttgcaaaa gtattacatc 1500
acgggggagg cagagggttt ccctgccaca gtctgactcg ag
                                                                  1542
```

```
<210> 19
<211> 511
<212> PRT
<213> Artificial Sequence
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<220>

<223> Description of Artificial Sequence: lexA-ER(alpha)EF fusion protein

Asp His Ile Ser Gln Thr Gly Met Pro Pro Thr Arg Ala Glu Ile Ala 20 25 30

Gln Arg Leu Gly Phe Arg Ser Pro Asn Ala Ala Glu Glu His Leu Lys Ala Leu Ala Arg Lys Gly Val Ile Glu Ile Val Ser Gly Ala Ser Arg Gly Ile Arg Leu Glu Glu Glu Glu Gly Leu Pro Leu Val. Gly Arg Val Ala Ala Gly Glu Pro Leu Leu Ala Gln His Ile Glu Gly His Tyr Gln Val Asp Pro Ser Leu Phe Lys Pro Asn Ala Asp Phe Leu Leu Arg Val Ser Gly Met Ser Met Lys Asp Ile Gly Ile Met Asp Gly Asp Leu Leu Ala Val His Lys Thr Gln Asp Val Arg Asn Gly Gln Val Val Val Ala Arg Ile Asp Asp Glu Val Thr Val Lys Arg Leu Lys Lys . Gln Gly Asn Lys Val Glu Leu Leu Pro Glu Asn Ser Glu Phe Lys Pro Ile Val Val Asp Leu Arg Gln Gln Ser Phe Thr Ile Glu Gly Leu Ala Val Gly Val Ile Arg Asn Gly Asp Trp Leu Glu Phe Lys Leu Glu Leu Gly Gly Ser Gly Met Ile Lys Arg Ser Lys Lys Asn Ser Leu Ala Leu Ser Leu Thr Ala Asp Gln Met Val Ser Ala Leu Leu Asp Ala Glu Pro Pro Ile Leu Tyr Ser Glu Tyr Asp Pro Thr Arg Pro Phe Ser Glu Ala Ser Met Met Gly Leu Leu Thr Asn Leu Ala Asp Arg Glu Leu Val His

Met Ile Asn Trp Ala Lys Arg Val Pro Gly Phe Val Asp Leu Thr Leu

His Asp Gln Val His Leu Leu Glu Cys Ala Trp Leu Glu Ile Leu Met 290 295 300

Ile Gly Leu Val Trp Arg Ser Met Glu His Pro Val Lys Leu Leu Phe 305 310 315 320

Ala Pro Asn Leu Leu Asp Arg Asn Gln Gly Lys Cys Val Glu Gly 325 330 . 335

Met Val Glu Ile Phe Asp Met Leu Leu Ala Thr Ser Ser Arg Phe Arg 340 345 350

Met Met Asn Leu Gln Gly Glu Glu Phe Val Cys Leu Lys Ser Ile Ile 355 360 365

Leu Leu Asn Ser Gly Val Tyr Thr Phe Leu Ser Ser Thr Leu Lys Ser 370 375 380

Leu Glu Glu Lys Asp His Ile His Arg Val Leu Asp Lys Ile Thr Asp 385 390 395 400

Thr Leu Ile His Leu Met Ala Lys Ala Gly Leu Thr Leu Gln Gln 405 410 415

His Gln Arg Leu Ala Gln Leu Leu Leu Ile Leu Ser His Ile Arg His 420 . 425 430

Met Ser Asn Lys Gly Met Glu His Leu Tyr Ser Met Lys Cys Lys Asn 435 440 445

Val Val Pro Leu Tyr Asp Leu Leu Leu Glu Met Leu Asp Ala His Arg
450 455 460

Leu His Ala Pro Thr Ser Arg Gly Gly Ala Ser Val Glu Glu Thr Asp 465 470 475 480

Gln Ser His Leu Ala Thr Ala Gly Ser Thr Ser Ser His Ser Leu Gln 485 490 495

Lys Tyr Tyr Ile Thr Gly Glu Ala Glu Gly Phe Pro Ala Thr Val $500 \hspace{1.5cm} 505 \hspace{1.5cm} 510$

<210> 20

<211> 5

<212> PRT

<213> Artificial Sequence

```
<220>
<223> Description of Artificial Sequence: protein
<220>
<221> UNSURE
<222> (2)..(3)
<223> X at any position can be any amino acid
<400> 20
Leu Xaa Xaa Leu Leu
<210> 21
<211> 4
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: endoplasmic
      reticulum localization signal
<400> 21
Lys Asp Glu Leu
<210> 22
<211> 5
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: BC loop
      sequence for polypeptide monobody in pFNB42B5F7
      library
<400> 22
Trp Tyr Gln Gly Arg
  1
<210> 23
<211> 5
<212> PRT
<213> Artificial Sequence
<220>
```

```
<223> Description of Artificial Sequence: BC loop
      sequence for polypeptide monobody in pFNB42B5F7
<400> 23
Pro Arg Thr Lys Gln
<210> 24
<211> 5
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: BC loop
      sequence for polypeptide monobody in pFNB42B5F7
      library
<400> 24
Val Arg Arg Pro Pro
  1
<210> 25
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pFNB42B5F7
      library
<400> 25
Gly Ile Leu Glu Met Leu Gln
  1
<210> 26
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pFNB42B5F7
```

library

```
<400> 26
 Arg Leu Arg Ala Gln Leu Val
   1
 <210> 27
 <211> 7
 <212> PRT
 <213> Artificial Sequence
 <220>
 <223> Description of Artificial Sequence: FG loop
       sequence for polypeptide monobody in pFNB42B5F7
       library
 <400> 27
 Pro Val Arg Val Leu Leu Arg
 <210> 28
 <211> 7
 <212> PRT
 <213> Artificial Sequence
 <220>
 <223> Description of Artificial Sequence: FG loop
       sequence for polypeptide monobody in pFNB42B5F7
       library
 <400> 28
 Arg Leu Arg Asp Leu Leu Gln
 <210> 29
 <211> 7
. <212> PRT
 <213> Artificial Sequence
<220>
 <223> Description of Artificial Sequence: FG loop
       sequence for polypeptide monobody in pFNB42B5F7
       library
<400> 29
Gly Leu Val Ser Leu Leu Arg
```

1 <210> 30 <211> 7 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pFNB42B5F7 library <400> 30 Arg Lys Val Val Trp Thr Gly <210> 31 <211> 7 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pFNB42B5F7 library <400> 31 Thr Ala Ala Ile Met Val Lys . 1 <210>. 32 <211> 5 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: consensus sequence <220> <221> UNSURE <222> (2)..(3)

31

<223> X at any position can be an amino acid

<400> 32

```
Leu Xaa Xaa Met Leu
<210> 33
<211> 5
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: sequence
      within helix 12 of estrogen receptor-alpha and
      estrogen receptor-beta
<400> 33
Leu Leu Glu Met Leu
  1
<210> 34
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: AB loop
      sequence for polypeptide monobody in pYT45AB7N
      library
<400> 34
Trp Thr Trp Val Leu Arg Glu
<210> 35
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: AB loop
      sequence for polypeptide monobody in pYT45AB7N
      library
<400> 35
Trp Val Leu Ile Thr Arg Ser
 1
```

```
<210> 36
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT45B3F7
      library
<400> 36
Leu Arg Leu Met Leu Ala Gly
<210> 37
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT45B3F7
      library
<400> 37
Ala Leu Val Glu Met Leu Arg
<210> 38
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT45B3F7
      library
<400> 38
Arg Leu Leu Trp Asn Ser Leu
. 1
<210> 39
<211> 7
```

```
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT45B3F7
      library
<400> 39
Arg Val Leu Met Thr Leu Leu
<210> 40
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT45B3F7
      library
<400> 40
Gly Leu Arg Arg Leu Leu Arg
<210> 41
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT45B3F7
      library
<400> 41
Gly Leu Arg Gln Met Leu Gly
  1
<210> 42
<211> 7
<212> PRT
<213> Artificial Sequence
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<220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT45B3F7 library <400> 42 Arg Val Leu His Ser Leu Leu <210> 43 <211> 7 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT45B3F7 library <400> 43 Arg Val Arg Asp Leu Leu Met 1 <210> 44 <211> 7 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT45B3F7 library <400> 44 Arg Val Met Asp Met Leu Leu 1 <210> 45 <211> 7 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT45B3F7

library <400> 45 Gly Ile Ala Glu Leu Leu Arg 1 5 <210> 46 <211> 8 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT45B3F7 library <400> 46 Arg Ile Leu Leu Asn Met Leu Thr 1 <210> 47 <211> 8 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT45B3F7 library <400> 47 Gly Gly Trp Leu Trp Cys Val Thr <210> 48 <211> 7 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT45B3F7 library

<400> 48

```
Thr Trp Val Val Arg Arg Val
<210> 49
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT45B3F7
      1ibrary
<400> 49
Thr Trp Val Arg Pro Asn Gln
                   5
<210> 50
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT45B3F7
      library
<400> 50
Arg Arg Val Pro Ile Trp Cys
<210> 51
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT45B3F7
      library
<400> 51
Arg Arg Val Tyr Asp Phe Leu
```

```
<210> 52
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT45B3F7
      library
<400> 52
Leu Arg Gln Met Leu Ala Asp
<210> 53
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT45B3F7
      library
<400> 53
Gly Leu Arg Met Leu Leu Arg
  1
<210> 54
<211> 16
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT47F16
      library
<400> 54
Ser Arg Arg Leu Val Glu His Leu Ala Gly Val Glu Val Gln Ala Leu
                                      10
 1
<210> 55
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<211> 16

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<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT47F16
      library
<400> 55
Leu Val Ala Arg Met Leu Asp Trp Ser Asp Gly Glu Glu Ala Ser Pro
<210> 56
<211> 16
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT47F16
      library
<400> 56
Gln Gly Lys Gly Arg Arg Gly Leu Val Leu Tyr Leu Leu Gly Ser
                                      10
                                                          15
<210> 57
<211> 16
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT47F16
      library
<400> 57
Arg Leu Arg Glu Leu Leu Ala Glu Ala Ala Gln Ala Ser Asp Gly Glu
  1 .
                                     10
                                                          15
<210> 58
<211> 16
<212> PRT
<213> Artificial Sequence
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<220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 library <400> 58 Leu Leu Leu Arg Val Gly Cys Gly Cys Arg Leu Val Gly Ser Val Leu 10 <210> 59 <211> 16 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 library <400> 59 Arg Leu Ser Ile Val Pro Cys Pro Ala Trp Ala Arg Leu Thr Val Leu 10 <210> 60 <211> 16 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 library <400> 60 Leu Leu Val Gly Leu Leu Leu Arg Gly Ala Arg Ser Gly Ser Thr 1 5 10 <210> 61 <211> 16 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop

sequence for polypeptide monobody in pYT47F16

library <400> 61 Leu Ile Tyr Gly Leu Leu Ser Gln Pro Glu Glu Arg Asp Glu Trp Arg 5 10 <210> 62 <211> 16 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 library <400> 62 Arg Ser Asp Gly Val Leu Leu Arg Leu Leu Ala Gly Gln Arg Asn Ala 1 5 <210> 63 <211> 16 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 library Trp Phe Asp His Glu Arg His Gly Met Leu Trp Gln Leu Leu Leu Arg 1 10 15 . <210> 64 <211> 16 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 library

<400> 64

```
Arg Leu Trp Cys Leu Leu Gln Arg Lys Gly Arg Asn Pro Ile Asp Met
                                      10
<210> 65
<211> 16
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT47F16
      library
<400> 65
Arg Val Phe Phe Gly Ile Gly Cys Arg Gly Gly Thr Gly Gly Asn
                                     10
                                                          15
<210> 66
<211> 16
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT47F16
      library
<400> 66
Arg Val Arg Phe Arg Cys Gly Gly Arg Asp Ala Ala Ser Gly Asp Gln
                                     10
<210> 67
<211> 16
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: FG loop
      sequence for polypeptide monobody in pYT47F16
      library
<400> 67
Leu Val Arg Phe Arg Val Val Asn Ser Ser Leu Cys Met Trp Ala Arg
```

10

<210> 68 <211> 16 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 library <400> 68 Leu Val Arg Leu Gly Val Ala Gly His Met Asp Ala Gly Ala Gly Arg 5 <210> 69 <211> 16 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 library <400> 69 Pro Ala Asp Gly Ser Glu Val Leu Arg Leu Val Lys Ile His Tyr Val 10 <210> 70 <211> 16 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 library <400> 70 Arg Leu Glu Tyr Gly Asp Val Ile Gly Ala Val Trp Trp Gly Arg Val 1 10 <210> 71

<211> 16

<212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 library <400> 71 Gln Gly Ala Ala Val Arg Thr Leu Val Ala Gly Gly Val Ala Ser 10 <210> 72 <211> 16 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 .. library <400> 72 Leu Glu Val Arg Val Ala Ala Gly Cys Ile Ala Gly Gly Gly Arg Arg 10 <210> 73 <211> 16 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: FG loop sequence for polypeptide monobody in pYT47F16 library <400> 73 Arg Leu Trp Arg Met Leu Ser Gly Glu Pro Ala Arg Val Asp His Glu 5 10